UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF PREVENTION,
PESTICIDES AND TOXIC SUBSTANCES

22/SEP/2000

MEMORANDUM

Subject: EPA File Symbol: 11556-REA Advantage Plus 9 for Cat,
11556-REO Advantage Plus 18 for Cats
DP Barcodes: D265762 and D265765
Case No: 068807, 068810
PC Code: 129099

From: Masih Hashim, Toxicologist MH
Technical Review Branch
Registration Division (7505C) SCR

To: Helene Daniel/Tina Levine, PM 04
Insecticide Rodenticide Branch
Registration Division (7505C)

Registrant: Bayer Corporation

ACTION REQUESTED: The Registrant requests a review of the companion animal safety study (MRID 45097001), Advantage Plus® 9 and 18 for cats with: 9.1% Imidacloprid w/w; 0.9% Pyriproyfen w/w (active ingredients). The compound was topically applied at 5 times (limit test) the recommended dose to groups of 6 male and 6 female cats. Animals were treated on study days 0, 7, 14, and 21.

COMMENTS/RECOMMENDATIONS:

The study MRID 45097001 was conducted at 5X the specified application rate; which demonstrates an adequate safety margin for adult cats. There was no repeated toxicological response in cats following exposure to the proposed formulation. Any such response is random and is seen in the earlier phase of study. This cat study has been classified as **Acceptable.**

The Products: 11556-REA and/or 11556-REO have Imidacloprid 9.15% and Pyriproxyfen 0.9%. However, the label states 9.1% Imidacloprid and 0.46% pyriproxyfen, having same formulation for both products.

This product has residential uses and an Oral LD_{50} value < 1500 mg/kg, which would require the Child Resistant Packaging.

Acute toxicology profile for the Product is as follows:

Acute Oral LD_{50}	III	acceptable
Acute Dermal LD_{50}	IV	acceptable
Acute Inhalation LC_{50}	IV	acceptable
Primary Eye Irritation	III	acceptable
Primary Dermal Irritation	IV	acceptable
Dermal Sensitization	neg	acceptable

Primary review of the Companion Animal Safety Study was conducted by an Agency Contractor, then revised by the Technical Review Branch. There are two labels, one for Advantage Plus 9 for Cats and the other for Advantage Plus 18 for Cats.

Following is the Executive Summary of the study:

In a companion animal safety study (MRID 45097001), Advantage Plus® 9 and 18 for cats (Active Ingredients: 9.1% Imidacloprid w/w; 0.9% Pyriproyfen w/w) was topically applied at dose volumes of 2.0 ml for cats weighing less than or equal to 9 lbs, and 4.0 mL for cats weighing greater than 9 lbs (5 times the recommended doses) to groups of 6 male and 6 female cats, 7 months to one year of age. Controls were dosed with the vehicle at volumes of 2.0 mL for cats weighing less than or equal to 9 lbs and 4.0 ml for cats weighing greater than 9 lbs (5.6 times the volume of vehicle in the recommended doses). Animals were treated on (study) days 0, 7, 14, and 21.

Treatment related clinical signs included transient salivation which ceased within 2 hours of treatment on day 0 (4 of 12 test animals and 1 of 12 vehicle control animals reported from licking the test material), and a rough hair coat appearance at the treatment site on all animals of both groups following treatment on days 14 and 21. None of the cats were observed salivating following the last 3 treatments. One animal from the test group had pruritis at one hour on day 21. There was vomiting by two cats in the test group on days 19 and 25, which did not occur in periods following the test (substance) applications. Vomiting may be associated with licking the test substance. This does not appear to be exposure related. There were loose stools from two cats in the control group. Additionally, one male from the control group exhibited inappetence on days 22-24 and was observed to be circling and unsteady at the p.m. observation period on day 23. The clinical chemistry and hematology findings from this cat on day 22 were consistent with hemoconcentration due to dehydration. Possible cause of these signs was not clear. His behavior and appetite were normal for the remainder of the study, as were his clinical pathology parameters. There were no other treatment related effects on hematology, coagulation or clinical chemistry parameters. There were no treatment related effects on body weight or food consumption, and there were no signs of irritation at the application sites.

Currently there is a product in the market with 9.1% Imidacloprid. The proposed product is adding 0.46% Pyriproxyfen to the current product. The added ingredient has low acute and chronic toxicity in mammalian species.

Any clinical signs on the study showed no consistent toxicological response. This study is classified as **Acceptable /Guideline** for a companion animal safety study (OPPTS 870.7200) in cats .

LABELING:

Date: 09/28/00 LABEL REVIEW SYSTEM

ID #: 011556-00126 Advantage Plus 9 for Cats

SIGNAL WORD: CAUTION

PRECAUTIONARY STATEMENTS:

Harmful if swallowed. Causes moderate eye irritation. Avoid contact with eyes or clothing.

STATEMENT OF PRACTICAL TREATMENT (SOPT):

IF SWALLOWED: Call a poison control center or doctor immediately for treatment advice. Have person sip a glass of water if able to swallow. Do not induce vomiting unless told to by a poison control center or doctor. Do not give anything by mouth to an unconscious person.

IF IN EYES: Hold eye open and rinse slowly and gently with water for 15-20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. Call a poison control center or doctor for treatment advice.

NOTE TO PHYSICIAN:

Note to PM/CRM/Registrant: The proposed label should contain a "Note to Physician." The following statements are suggested types of information that may be included, if applicable:

- technical information on symptomatology;
- use of supportive treatments to maintain life functions;
- medicine that will counteract the specific physiological effects of the pesticide;
- company telephone number to specific medical personnel who can provide specialized medical advice.

USER SAFETY RECOMMENDATION

Wash hands before eating, drinking, chewing gum, using tobacco or using the toilet.

ID #: 011556-00129 Advantage Plus 18 for Cats

SIGNAL WORD: CAUTION

PRECAUTIONARY STATEMENTS:

Harmful if swallowed. Causes moderate eye irritation. Avoid contact with eyes or clothing.

STATEMENT OF PRACTICAL TREATMENT (SOPT):

IF SWALLOWED: Call a poison control center or doctor immediately for treatment advice. Have person sip a glass of water if able to swallow. Do not induce vomiting unless told to by a poison control center or doctor. Do not give anything by mouth to an unconscious person.

IF IN EYES: Hold eye open and rinse slowly and gently with water for 15-20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. Call a poison control center or doctor for treatment advice.

NOTE TO PHYSICIAN:

Note to PM/CRM/Registrant: The proposed label should contain a "Note to Physician." The following statements are suggested types of information that may be included, if applicable:

- technical information on symptomatology;
- use of supportive treatments to maintain life functions;
- medicine that will counteract the specific physiological effects of the pesticide;
- company telephone number to specific medical personnel who can provide specialized medical advice.

USER SAFETY RECOMMENDATIONS: Wash hands before eating, drinking, chewing gum, using tobacco or using the toilet.

DATA EVALUATION REPORT

ADVANTAGE PLUS® 9 AND 18 FOR CATS
[9.1% Imidacloprid with 0.9% Pyriproxyfen Spot-on Formulation]

STUDY TYPE: Companion Animal Safety - Cat (OPPTS 870.7200)
MRID 45097001

Prepared for

Registration Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831

Primary Reviewer: Donna L. Fefee, D.V.M..	Signature: Date:	Robert H. Ross for D.L. Fefee AUG 14 2000
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast AUG 14 2000
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross AUG 14 2000
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson AUG 14 2000

Disclaimer

This review may have been altered subsequent to the contractors signatures above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC, for the U.S. Department of Energy under contract number DE-AC05-00OR22725.

DATA EVALUATION RECORD

STUDY TYPE: Companion Animal Safety/Cats[OPPTS 870.7200]

EPA I.D. NUMBERS: DP BARCODES: D265762, D265765; MRID NUMBER: 45097001

TEST MATERIAL: Advantage Plus® 9 and 18 for Cats

STUDY NUMBER: 75122 (150.853)

TESTING FACILITY: Bayer Corporation, Agriculture Division, Animal Health, DeSoto Research Facility, 35040 West 87th Street, Building Number 20, DeSoto, Kansas 66018.

SPONSOR: Bayer Corporation, Agriculture Division, Animal Health

TITLE OF REPORT: Evaluation of the general safety of 9.1% Imidacloprid with 0.9% pyriproxyfen spot-on formulation in the target species, adult cats.

AUTHOR: A.S. Abraham

REPORT ISSUED: April 4, 2000

EXECUTIVE SUMMARY:.

In a companion animal safety study (MRID 45097001), Advantage Plus® 9 and 18 for cats (Active Ingredients: 9.1% Imidacloprid w/w; 0.9% Pyriproyfen w/w) was topically applied at dose volumes of 2.0 ml for cats weighing less than or equal to 9 lbs, and 4.0 mL for cats weighing greater than 9 lbs (5 times the recommended doses) to groups of 6 male and 6 female cats, 7 months to one year of age. Controls were dosed with the vehicle at volumes of 2.0 mL for cats weighing less than or equal to 9 lbs and 4.0 ml for cats weighing greater than 9 lbs (5.6 times the volume of vehicle in the recommended doses). Animals were treated on (study) days 0, 7, 14, and 21.

Treatment related clinical signs included transient salivation which ceased within 2 hours of treatment on day 0 (4 of 12 test animals and 1 of 12 vehicle control animals reported from licking the test material), and a rough hair coat appearance at the treatment site on all animals of both groups following treatment on days 14 and 21. None of the cats were observed salivating following the last 3 treatments. One animal from the test group had pruritis at one hour on day 21. There was vomiting by two cats in the test group on days 19 and 25, which did not occur in periods following the test (substance) applications. Vomiting may be associated with licking the test substance. This does not appear to be exposure related. There were loose stools from two cats in the control group. Additionally, one male from the control group exhibited inappetence on days 22-24 and was observed to be circling and unsteady at the p.m. observation period on day 23. The clinical chemistry and hematology findings from this cat on day 22 were consistent with hemoconcentration due to dehydration. Possible cause of these signs was not

clear. His behavior and appetite were normal for the remainder of the study, as were his clinical pathology parameters. There were no other treatment related effects on hematology, coagulation or clinical chemistry parameters. There were no treatment related effects on body weight or food consumption, and there were no signs of irritation at the application sites.

Currently there is a product in the market with 9.1% Imidacloprid. The proposed product is adding 0.46% Pyriproxyfen to the current product. The added ingredient has low acute and chronic toxicity in mammalian species.

Any clinical signs on the study showed no consistent toxicological response. This study is classified as **Acceptable /Guideline** for a companion animal safety study (OPPTS 870.7200) in cats .

I. MATERIALS

A. Test material

9.1% Imidacloprid with 0.9% Pyriproxyfen (w/w) Spot-on Formulation (Advantage Plus® 9, and 18 for Cats)
Description: not provided
Lot No.: 99-901-66
Active Ingredients: Imidacloprid, 9.1% (w/w); Pyriproxyfen, 0.9% (w/w)
Storage Conditions: in the dark in a closed cabinet at room temperature

B. Administration: Topical (spot-on)

C. Vehicle and/or positive control

The control animals received the vehicle.

D. Test animals

Species: Cat
Breed: Domestic shorthair
Age and weight at study initiation: 7-12 months; males: 3.8-5.1 kg., females: 2.4-3.1 kg
Source: Liberty Research Inc., 170 Route 17C, P.O. Box 107, Waverly, New York
Housing: Individually in cages with approximately 7.5 square feet of floor space
Diet: Commercial feed purchased from Harlan Teklad, Madison, Wisconsin, once daily
Water: Tap water, *ad libitum*
Environmental conditions:
Temperature: not reported
Humidity: not reported
Air changes: not reported
Photoperiod: not reported
Acclimation period: 14 days

II. STUDY DESIGN

A. In life dates: start: August 11, 1999; end: September 21, 1999

B. Animal assignment/ Dosage and Administration

Cats were assigned to the groups in Table 1 using stratified blocked randomization according to weight. Group 1 received the test substance at 5X the label specified use volume, and group 2 received the vehicle without the two active ingredients at a volume equivalent to the 5X use rate volume of the test substance. The dose volume was 2.0 mL for cats of either group weighing less than or equal to 4.1 kg (9 lbs) and 4.0 mL for cats of either group weighing greater than 4.1 kg. Treatments were applied on the back, from the back of the head to the shoulder. Animals were dosed on Study Days 0, 7, 14, and 21. Dose volumes for treatments on study days 0 and 7 were determined using body weights from study day -1, and dose volumes for treatments on study days 14 and 21 were determined using body weights from study day 13.

TABLE 1. Study design

Group	Number of animals		Dose volume (mL)/multiple of recommended dose		Number of applications[a]
	Male	Female	Body weight≤9 lbs	Body weight>9 lbs	
1. Test substance	6	6	2.0 /5X	4.0/5X	4
2. Vehicle control	6	6	2.0 /5.6X	4.0/5.6X	4

Data taken from pp. 12-13, 16-17, MRID 45097001.

[a] Treatments were given on study days 0, 7, 14, and 21.

C. Dose selection rationale

The study was conducted as a limit test using 5 times the label specified, the recommended dose volume. The product was dosed according to weight in the following pre-measured dose volumes: 0.4 mL for cats weighing ≤9 lbs (4.1 kg) and 0.8 mL for cats weighing > 9 lbs. (4.1kg). The vehicle control group received the vehicle at dose volumes equal to 5 times the recommended use volumes of the test substance, which are equivalent to 5.6 times the usual use volumes of the vehicle. The product is intended for once a month use; however, the label states that "if re-treatment becomes necessary earlier than four weeks, do not re-treat more than once weekly." The study therefore included repeated treatments at weekly intervals for a total of four treatments.

D. Experimental design

The cats were observed daily during study days -14 through -1 (the acclimation period), and twice daily during study days 0 through 37 except on dosing days, when the animals were observed once prior to dosing and 4 times, approximately 1 hour apart, following dosing. These observations included evaluation of the "clinical condition" of the eyes,

appetite, feces, respiration, behavior changes, locomotion and musculature, skin, including dermal irritation, and any signs of vomiting. Physical examinations were conducted prior to the acclimation period and on study days -1 and 37. Body weights were recorded on study days -14, -7, -1, 13, 28, and 37. Food consumption was evaluated once daily during the acclimation period and twice daily during the study except on dosing days, when it was evaluated 5 times; in all cases, a daily summary of food consumption was also made. The amount of food consumed was estimated visually and scored as 1, 2, or 3, indicating, respectively, that greater than or equal to 75%, 25-75%, or less than 25% of the food was consumed; however, the quantity of food the animals were given was not provided in the report.

E. Pathological parameters

Baseline blood samples were collected on study days -7 and -1, and post-treatment blood samples were collected on study days 1, 22, and 37. The report did not mention the venipuncture sites used or whether the animals were fasted overnight prior to blood collection. Due to clotting of some blood samples, it was necessary to collect and test additional samples. Where necessary, study day 1 sampling and testing were repeated on study 8, study day 22 blood sampling and testing were repeated on day 23, and study day 37 blood sampling and testing were repeated on study days 41 and 42. The CHECKED (X) parameters were examined.

a. Hematology

X		X	
X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*	X	Mean corpusc. HGB conc.(MCHC)*
X	Erythrocyte count (RBC)*	X	Mean corpusc. volume (MCV)*
X	Platelet count		Reticulocyte count
	Blood clotting measurements		
	(Thromboplastin time)		
	(Clotting time)		
X	(Prothrombin time)*		
X	(Activated partial thromboplastin time)*		
	Erythrocyte morphology		

*Recommended in OPPTS 870.7200 Guidelines.

b. Clinical chemistry

X	ELECTROLYTES	X	OTHER
X	Calcium*	X	Albumin*
X	Chloride*	X	Blood creatinine*
	Magnesium	X	Blood urea nitrogen*
X	Phosphorus*		Total Cholesterol
X	Potassium*	X	Globulin*
X	Sodium*	X	Glucose*
		X	Total and direct bilirubin*
	ENZYMES	X	Total serum protein (TP)*
X	Alkaline phosphatase(ALK)*		Triglycerides
	Cholinesterase(ChE)		Serum protein electrophoresis
	Creatine kinase		Albumin/Globulin ratio
	Lactic acid dehydrogenase(LDH)	X	Calcium/phosphorus ratio
X	Serum alanine amino- transferase (also SGPT)*	X	Blood urea nitrogen/creatinine ratio
X	Serum aspartate amino- transferase(also SGOT)*	X	Sodium/potassium ratio
	Gamma glutamyl transferase(GGT)		
	Amylase		
	Glutamate dehydrogenase		

*Recommended in OPPTS 870.7200 Guidelines.

F Statistics

Baseline clinical pathology values were calculated for each animal by averaging the pre-treatment measurements, and study day -1 body weights were used as baseline. Body weight data were analyzed by first comparing the baseline body weights of the two groups by sex with a two-sample t-test. Body weight changes from baseline were calculated for each post-treatment day (study days 13, 20, 28, and 37), and body weight changes were then analyzed by sex with a repeated measure analysis of covariance including terms for Group, Animal (random), Day, and Group*Day interaction with baseline body weight as the covariate. Clinical pathology data were analyzed using a multivariate repeated measures ANOVA, including terms for Group, Sex, Animal (random), Day, and Group*Day interaction. If the Sex effect was statistically significant at the 0.10 level, the data were analyzed by sex with a multivariate repeated measures ANOVA including terms for Group, Animal (random), Day, and Group*Day interaction. If the Group*Day interaction was statistically significant at the 0.10 level, the data were graphed to investigate the nature of the Group by Day interaction, and the data for each study day were compared with normal ranges.

G. Disposition of animals

Not reported.

H. Compliance

Signed and dated Quality Assurance, Data Confidentiality, and Good Laboratory Practice Statements were present.

III. RESULTS

A. Exposure levels

Each 1.0 mL of the product contained 100 mg of imidacloprid and 10 mg of pyriproxyfen. For thirty days efficacy, the minimum desired efficacious dose for imidacloprid is 10 mg/kg, and the desired minimum efficacious dose for pyriproxyfen is 0.5 mg/kg. In terms of desired minimum efficacious doses in mg/kg, the exaggerated doses used in the study ranged from 4.92X to 9.57X for imidacloprid and 9.84X to 19.13X for pyriproxyfen.

B. Mortality

There were no deaths during the study.

C. Clinical signs

Clinical observations are summarized in Table 2. Within an hour following the initial treatment on study day 0, four cats in the test substance group and one cat in the vehicle control group were observed to be salivating, and at two hours post dosing all had recovered. The study author stated that "all the cats that salivated were due to licking the test material." Two males in the test substance group vomited, one on study day 19 and the other on study day 25. Two males in the vehicle control group had loose stools, one on study days 18 and 20, and the other on study day 33. One cat in the vehicle control group was observed to be unsteady and circling during the p.m. observation period on study day 23. By the a.m. observation period on study day 24, he had recovered but exhibited a rough hair coat condition at the application site. No mention was made of the study veterinarian examining this animal during the time he was exhibiting symptoms. On study days 14 and 21, rough hair coat was noted at the application sites of all cats of both groups 1-4 hours post dosing, and, on study day 21, one female from the test substance group was pruritic at one hour after dosing, with recovery by 2 hours after dosing. There were no observations of erythema, edema, or alopecia at the application sites. One cat in the vehicle control group exhibited ocular discharge on study day -9 (during acclimation) and study day 16; since this condition occurred both before and after treatment, it is unlikely to be treatment related.

TABLE 2. Clinical observations of cats treated with Imidacloprid/Pyriproxyfen Spot-on Formulation		
Treatment group	**Day**	**Observation**
1. Test substance	0	Salivation in 3 males and 1 female within an hour of dosing
	14	Rough hair coat condition at the dose site in 6 males and 6 females at the post dosing observation periods at 1-4 hours post dosing
	16	Ocular discharge in one female cat at the a.m. observation period[a]
	19	Vomiting by one male cat (#762) at the a.m. observation period
	21	Rough hair coat condition at the dose site in 6 males and 6 females at the post dosing observation periods at 1-4 hours post dosing; Pruritis in one female at 1 hour post dosing
	25	Vomiting by one male cat (#758) at the p.m. observation period
2. Vehicle control	0	Salivation in one female within an hour of dosing
	14	Rough hair coat condition at the dose site in 6 males and 6 females at the post dosing observation periods at 1-4 hours post dosing
	18	Loose stools in one male cat (#764)
	20	Loose stools in one male cat (#764)
	21	Rough hair coat condition at the dose site in 6 males and 6 females at the post dosing observation periods at 1-4 hours post dosing
	23	One male (#744) was circling and unsteady at the p.m. observation
	24	Rough hair coat condition at the dose site in one male (#744)
	33	Loose stools in one male cat (#749)

Data taken from Tables 6A and 6B, pp. 34-35, MRID 45097001.
[a] This cat also exhibited ocular discharge on study day -9, during acclimation.

D. Bodyweight and weight gain

Individual body weights of the cats fluctuated throughout the acclimation and treatment periods, with no consistent pattern being observed. Mean body weights of both groups, with the sexes both combined and separated, increased at each consecutive weighing from study day -1 to study day 37. There were no significant differences between groups for post-treatment changes in body weight

E. Food consumption

All of the cats on the study generally consumed greater than or equal to 75% of their food. One female (#750) in the test substance group consumed between 25 and 75% of her food on study days -13, 12, 15-17, 19, and 30-33. One female (#751) in the vehicle control group consumed between 25 and 75% of her food on study days 13, 14, 16-18, 31-32. Three additional cats from the test substance group and one cat from the vehicle control group consumed 25-75% of their food for 1-3 days. Cat number 744 from the vehicle control group, who was observed to be circling and unsteady on study day 23, consumed less than 25% of his food on study day 22 and 25-75% of his food on study days 23 and 24. All of the previously mentioned animals consumed greater than or equal

to 75% of their food during the remainder of the pre-treatment and treatment intervals. The study report did not include the quantities of food the cats were fed.

F. Hematology

Nine of the samples submitted for complete blood count and three of the samples submitted for coagulation measurements were could not be analyzed due to clotting. Additional samples were collected and tested as follows: study day 1 sampling and testing were repeated on study day 8; study day 22 sampling and testing were repeated on study day 23; and study day 37 sampling and testing were repeated on study days 41 and 42. Statistical analyses were conducted both with and without the data from the repeated tests, and wherever statistical significance was present without data from the repeated tests, it was also present when the data from the repeated tests was included. The individual results from these repeated tests were omitted from the study report although these data were included in calculating means and standard deviations. Statistically significant ($p<0.10$) Group by Day interactions were found for the following parameters: platelet counts for males, erythrocyte counts for females, and activated partial thromboplastin times, leukocyte, eosinophil, and lymphocyte counts, and mean corpuscular volumes for the pooled sexes. These values are given in Table 3. These values were all within pre-treatment ranges, and the changes were considered to be spontaneous in nature and unrelated to treatment.

On study day 22, cat #744 (the cat that was observed to be circling and unsteady on study day 23) exhibited an increased hematocrit, erythrocyte count, and hemoglobin concentration. These values were all outside the pre-treatment ranges for these parameters. (See G. Clinical chemistry below.)

TABLE 3. Hematology and coagulation parameters in cats treated with Imidacloprid/Pyriproxyfen Spot-on Formulation which exhibited statistically significant (p<0.10)Changes, Group by Day interactions [a]								
Parameter	1. Test substance				2. Controls			
	Baseline [b]	Day 1[c]	Day 22[d]	Day 37[e]	Baseline	Day 1[f]	Day 22[g]	Day 37[h]
Males								
Platelets ($10^6/\mu L$)	0.33	0.26	0.34	0.39	0.30	0.29	0.34	0.33
Females								
Erythrocytes ($10^6/\mu L$)	9.71	8.73	8.48	8.85	8.46	7.86	8.35	8.65
Pooled Sexes								
Activated partial thromboplastin time (sec)	12.68	13.03	12.21	12.28	13.53	12.80	12.94	13.34
MCV (fl)	41.33	41.19	41.34	41.47	42.81	43.08	43.27	42.85
Leukocytes ($10^3\mu L$)	15.05	14.84	11.44	11.99	13.74	15.07	13.12	10.24
Eosinophils ($10^3/\mu L$)	0.81	1.10	0.43	0.43	0.93	0.90	0.65	0.39
Lymphocytes ($10^3/\mu L$)	6.22	5.44	4.04	5.13	4.68	4.63	3.87	3.26

Data taken from Tables III.1 and III.2, pp. 81-82, MRID 45097001

[a] Mean values

[b] Baseline values are the means of results from study days -7 and -1.

[c] Means for complete blood count (CBC) parameters include data from Day 8 for one female.

[d] Means for CBC parameters include data from Day 23 for one male.

[e] Means for CBC parameters include data from Day 40 or 41 for two males and two females.

[f] Means for CBC parameters include data from Day 8 for two females and one male. Means for coagulation parameters include data from Day 8 for one female.

[g] Means for CBC parameters include data from Day 23 for one female

[h] Means for coagulation parameters include data from Day 40 or 41 for one male.

G. Clinical chemistry

Statistically significant (p<0.10) Group by Day interactions were found for the following clinical chemistry parameters: alkaline phosphatase and BUN/Creatinine ratio in males. These values were within the pretreatment ranges, and the changes were considered to be spontaneous in nature and unrelated to treatment.

On study day 22, cat #744 (the cat that was observed to be circling and unsteady on study day 23) exhibited increased sodium concentration and total protein; both these values exceeded the pre-treatment ranges. There were also slight increases in his BUN on study

days 1 and 37 and creatinine. These findings in conjunction with those mentioned above (see F. Hematology) are consistent with hemoconcentration due to dehydration.

TABLE 4. Clinical chemistry parameters in cats treated with Imidacloprid/Pyriproxyfen Spot-on Formulation which exhibited statistically significant ($p<0.10$) Group by Day interactions [a]

Parameter	1. Test substance				2. Controls			
	Baseline	Day 1	Day 22	Day 37	Baseline	Day 1	Day 22	Day 37
Males								
Alkaline phosphatase activity (u/L)	52.92	48.33	48.50	45.50	48.92	52.00	53.17	39.00
BUN/creatinine ratio	18.86	18.43	17.56	19.60	18.29	18.33	19.77	18.73

Data taken from Tables III.1, p. 81, MRID 45097001.
[a] Mean values

H. Necropsy findings

Necropsies and histopathological examinations were not performed, as all animals survived until termination of the study, and no animals displayed clinical signs which were considered to warrant necropsy.

IV. DISCUSSION

A. Treatment related clinical signs included salivation and a rough hair coat appearance at the treatment site. Four cats from the test group and one from the control group exhibited salivation after the first treatment. Salivation was first noted within an hour of dosing and ended before the 2 hour post-treatment observation period. The study author attributed the salivation to the animals licking the test substance or vehicle. A rough hair coat appearance at the application site was noted for all cats in both groups on study days 14 and 21 during all post-treatment observations, and one animal from the test group was pruritic at one hour post dosing on study day 21. Clinical signs that may have been related to treatment included vomiting by two cats in the test group and loose stools from two cats in the (vehicle) control group. Additionally, one male from the control group exhibited inappetence on study days 22-24 and was observed to be circling and unsteady at the p.m. observation period on day 23. Clinical chemistry and hematology findings from this cat on day 22 were consistent with hemoconcentration due to dehydration (elevated hematocrit, erythrocyte count, hemoglobin, BUN, creatinine, sodium, and total protein), but since this cat was apparently not given a physical examination, it is unknown whether he was clinically dehydrated, febrile, or presenting neurological deficits. His behavior and appetite were normal for the remainder of the study, as were his clinical pathology parameters. There were no other treatment related effects on hematology, coagulation, and clinical chemistry parameters. There were no

treatment related effects on body weights or food consumption, and there was no evidence of irritation at the application sites. Since the cat that exhibited inappetence, circling, and unsteadiness following the fourth treatment was in the control group, these clinical signs were clearly not due to toxic effects from the active ingredients of the product; however, they may represent toxic effects from an ingredient in the vehicle. The animal was not examined more closely. However, the signs were transient, and the animal apparently recovered with no clinical signs for the remainder of the study and no abnormal physical examination findings on study day 37. The guideline only requires a single re-treatment with observation of the animals to continue only 14 days beyond treatment if no clinical signs of toxicity are noted. As the clinical signs in question did not occur until day 22, (exactly 15 days after the second treatment and were exhibited by an animal from the vehicle control group), they are not considered to be a treatment related.

B. Study deficiencies

An animal on the study exhibited clinical signs which needed a thorough examination to determine the possible cause of such clinical signs.

Due to clotting of some blood samples, it was necessary to collect and test additional samples. Where necessary, study day 1 sampling and testing were repeated on study 8, study day 22 blood sampling and testing were repeated on day 23, and study day 37 blood sampling and testing were repeated on study days 41 and 42. Individual data from the repeated tests were not included in the study report. The guideline requires clinical pathology testing 24 hours after treatment with additional assessment on day 7 if the day 1 results are altered. It is unclear why the tests on day 1 were not repeated until day 8.